In This Issue

X-ray Footprints on ClpA

PAGE 1157

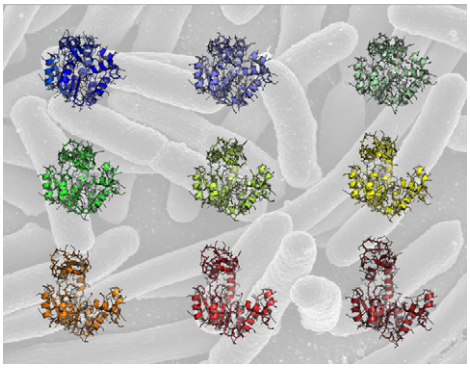
Changes in solvent accessibility upon ATPγS-induced hexamerization of ClpA, the chaperone component of the ATP-dependent protease ClpAP, were investigated by Bohon et al. using synchrotron protein footprinting. In this technique, large quantities of hydroxyl radicals are generated by direct irradiation of a protein solution, leading to oxidative modification of protein side chains of solvent-exposed protein regions. The results obtained for ATPγS-bound ClpA were compared with values derived from models constructed using the crystal structure of the ADP-bound monomer. Observed differences are attributed to distinct conformational states induced by different bound nucleotides and provide the first direct experimental support for a nucleotide-dependent conformational change previously proposed to mediate substrate translocation.

Lesion-Containing DNA Duplexes Easier to See

PAGE 1166

The constant attack on DNA by endogenous and exogenous agents gives rise to nucleobase modifications that cause mutations, which can lead to cancer. Visualizing the effects of these lesions on the structure of duplex DNA is the key to understanding their biologic consequences. The most definitive method of obtaining such structures, X-ray crystallography, is troublesome to employ owing to the difficulty of obtaining diffraction-quality crystals of DNA. Bowman et al. present an innovative crystallization system that uses a protein scaffold to mediate the crystallization of duplex DNA, which facilitated the rapid structure determination of several lesion-containing DNA duplexes.

TEE-REX Tackles Adenylate Kinase



PAGE 1175

Elucidating the pathways and mechanisms of protein conformational dynamics is a challenging task for both experimental and computational approaches, in particular, fully atomistic molecular dynamics (MD) simulations. Using the highly efficient TEE-REX algorithm, Kubitzki and de Groot successfully simulated the large conformational transition of *E. coli* adenylate kinase, a ubiquitous enzyme that plays a key role in intracellular energy maintenance by controlling ATP levels. The simulations reveal a detailed picture of a two-phasic transition pathway and identify the key residues stabilizing conformational substates of this highly flexible enzyme. (Figure credits: Kubitzki and de Groot)

Cyanovirin-N Mutant Doesn't Swap Domains

PAGE 1183

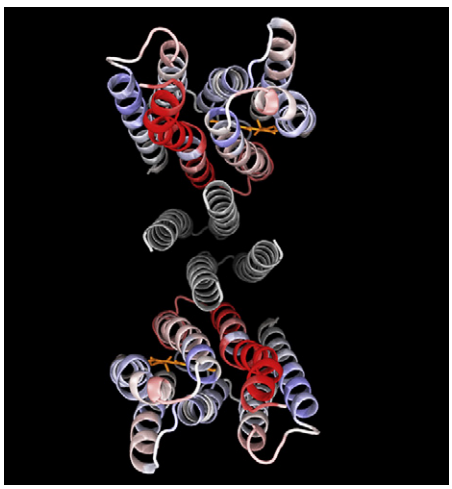
Potent anti-HIV activity was observed for the cyanobacterial protein CV-N, involving binding to high-mannose oligosaccharides on gp120. In the present mutant, Matei et al. abolish one of the two sugar binding sites and find the protein to be monomeric in solution and in the crystal, in contrast to wild-type CV-N that forms a domain-swapped dimer in the crystal. Binding of mutant CV-N to Man-9 is in the micromolar range, contrasting the situation for wild-type CV-N, where apparent high affinity through cross-linking was observed. Interestingly, the mutant is also completely inactive against HIV, suggesting that the antiviral activity is related to the multisite nature of the interaction with glycosylated gp120.

Type II Cadherin Adhesive Domain Does Swap

PAGE 1195

Cadherins are cellular adhesion molecules which interact through dimerization of N-terminal domains by exchange of N-terminal β-strands with symmetric insertion of conserved tryptophan indole side chains from one molecule into the hydrophobic acceptor pocket of the partner molecule. In the present work, Miloushev et al. apply nuclear magnetic resonance (NMR) spectroscopy to investigate the N-terminal domain of a classical Type II cadherin. The N-terminal β-strand is found to be dynamic in the monomer state, sampling exposed conformations and hence suggesting a possible mechanism to dimer formation.

May the Force Be with Sensory Rhodopsin



PAGE 1206

Membrane proteins are involved in virtually every biological process. To increase their functional repertoire, membrane proteins form supramolecular assemblies. Sensory rhodopsin II (SRII) can either adsorb light to pump ions across the cell membrane or associate with a transducer protein (HtrII) to mediate a photophobic response to avoid photo-oxidative damage. How does the transducer cause SRII to function differently? Using single-molecule force spectroscopy, Cisneros et al. characterize the intramolecular interactions within SRII upon HtrII binding. These results provide unique insights into molecular mechanisms, "priming" the complex for signaling and guiding the receptor towards transmitting light-activated structural changes to its cognate transducer. (Figure credit: Cisneros et al.)

Sliding and Binding Hybrid

PAGE 1214

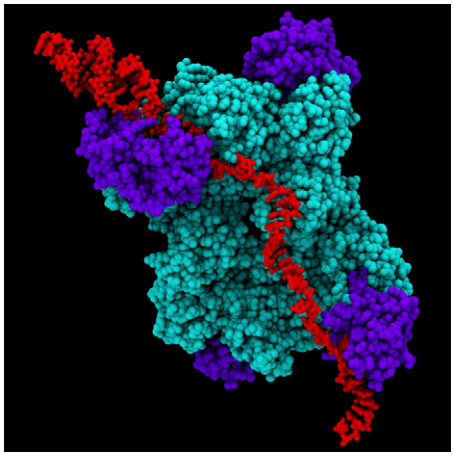
Human cytomegalovirus causes severe diseases in immunocompromised individuals. Production of the virus depends upon a processivity factor, UL44, which stimulates long-chain DNA synthesis by reducing dissociation of DNA polymerase from DNA. UL44 is also a potential target for new antiviral drugs. Komazin-Meredith et al. used computational and experimental approaches to develop the first atomic-level model of the bound DNA/UL44 structure. It shows that a dimer of U44 not only binds DNA directly, like other herpesvirus processivity factors, but encircles DNA like ring-shaped sliding clamp processivity factors. The results suggest mechanisms of UL44/DNA binding and the motion of UL44 on DNA during replication.

Biological "Nanobrick": S-Layer Protein

PAGE 1226

Surface layers (S-layers) comprise the outermost cell envelope component of most archaea and bacteria, representing up to 10% of the total protein content of the cell. The prominent feature of S-layer proteins is their ability to self-assemble on the cellular surface. Pavkov et al. now combine X-ray crystallography and small angle X-ray scattering to show that the structure of the bacterial S-layer protein SbsC from *Geobacillus stearothermophilus* is a very elongated and flexible molecule. Binding studies together with the structure provide insight into the processes of S-layer attachment to the underlying cell wall.

RNA in Tight Embrace



PAGE 1238

RNase E is an essential endoribonuclease in *Escherichia coli* that cleaves RNA internally during transcript turnover and processing of structured RNA precursors. However, the catalytic activity of the enzyme is sensitive to the identity of the chemical group at the distant 5' position of the RNA substrate. Crystal structure, reported by Koslover et al., reveals that the binding of the 5' end of the RNA in a sensor pocket favors a large conformational change that organizes the active site. Observed flexibility in the quaternary structure may allow the protein to mold to structured RNA substrates. (Figure credits: Koslover et al.)

Cation-π Interaction, "Methyl-Methyl" Switching, and Dynamic Regulation

PAGE 1245

Posttranslational modifications of histone proteins present unique chemical marks on the nucleosome surface that serve as binding sites for effector proteins. Trimethylation of lysine 4 of histone H3 has recently been shown to lead to the binding of basal transcription factor TFIID subunit TAF3, providing an explanation for its correlation with gene activation. Van Ingen et al. explore the structural basis of this interaction. They show that the plant homeodomain (PHD) finger of TAF3 features a unique structure rearrangement, underscoring the requirement for cation-π interactions in the binding. Interference by asymmetric dimethylation of arginine 2 suggests a "methyl-methyl" switch to dynamically regulate TFIID–promoter association.

Light the MATCHSTIX

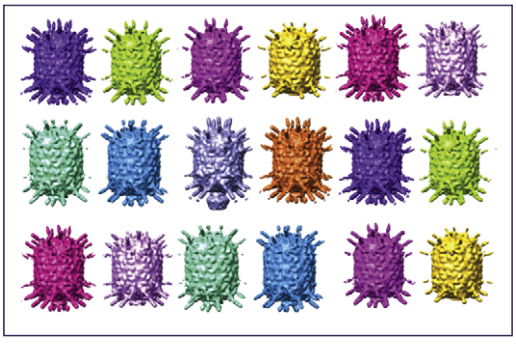
PAGE 1257

The assembly of tertiary folds from secondary structural elements is often the bottleneck in protein folding simulation and structure prediction. Here, Wu et al. present a fast assembly method for helical proteins. The algorithm, MATCHSTIX, involves rigid-body hydrophobic matching of two helical fragments absent the connecting loop, followed by loop closure and selection of a representative set of conformations for the next iteration, adding one helix at a time. Simulation on a set of 28 helical proteins shows that this rapid maximization of hydrophobic interactions can lead to small ensembles of highly native-like structures. It may be useful for protein structure prediction.

Cranking Viral DNA Packaging Motor

PAGE 1267

Many viruses and bacteriophages package their genomes into preformed protein shells. The process of genome encapsidation is remarkable considering the entropic, electrostatic, and bending energies of DNA that must be overcome in order to package DNA to near-crystalline density. Here, Morais et al. report a series of cryoEM reconstructions which characterize the different components of the bacteriophage ϕ29 DNA packaging motor. By comparing reconstructions of motor-intact particles to reconstructions of particles engineered to lack particular motor components, it was possible to delineate the molecular envelopes of individual motor components and suggest a mechanism for translocation of the genomic DNA. (Figure adapted from Morais et al.)



Tetrameric Structure of a Serine Integrase Catalytic Domain

PAGE 1275

Serine integrases have recently emerged as powerful new chromosome engineering tools in a variety of organisms and show promise for therapeutic use in human cells. Yuan et al. report the crystal structure and solution properties of the catalytic domain from bacteriophage TP901-1 integrase. The protein is a dimer in solution, but crystallizes as a tetramer that represents a unique intermediate on the recombination pathway that is shared within the serine recombinase superfamily. This work marks an important first step in relating the serine integrases to the well-studied resolvase/invertase enzymes and in developing a structural framework for understanding unique aspects of the integrase recombination pathway.

